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A harmonized PBPK model of hexavalent chromium in rats and mice

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The views expressed are those of the authors, and do not necessarily represent the views or policies of the U.S. EPA.

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Overview

- Integrated Risk Information System (IRIS) background
- Hexavalent chromium toxicity and carcinogenicity
  - –Toxicokinetics in the gastrointestinal (GI) tract
- Adaptation of toxicokinetic models
  - –Updated kinetic model for GI metabolism
  - –Revisions to whole-body model assumptions
- Application to National Toxicology Program data
- Remaining issues, Q&A

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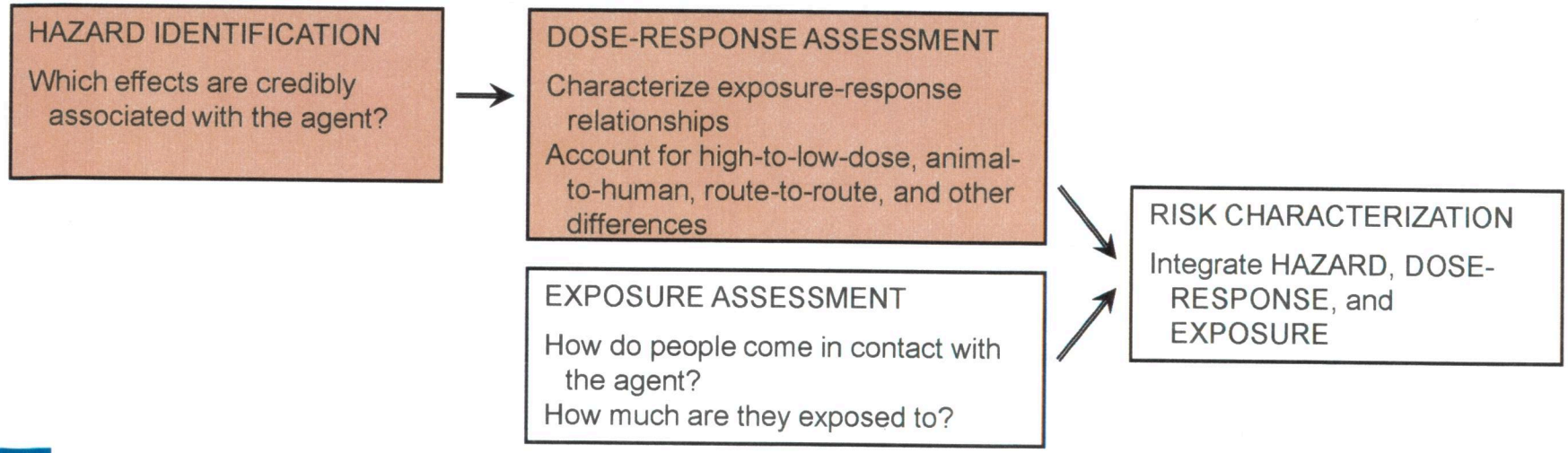
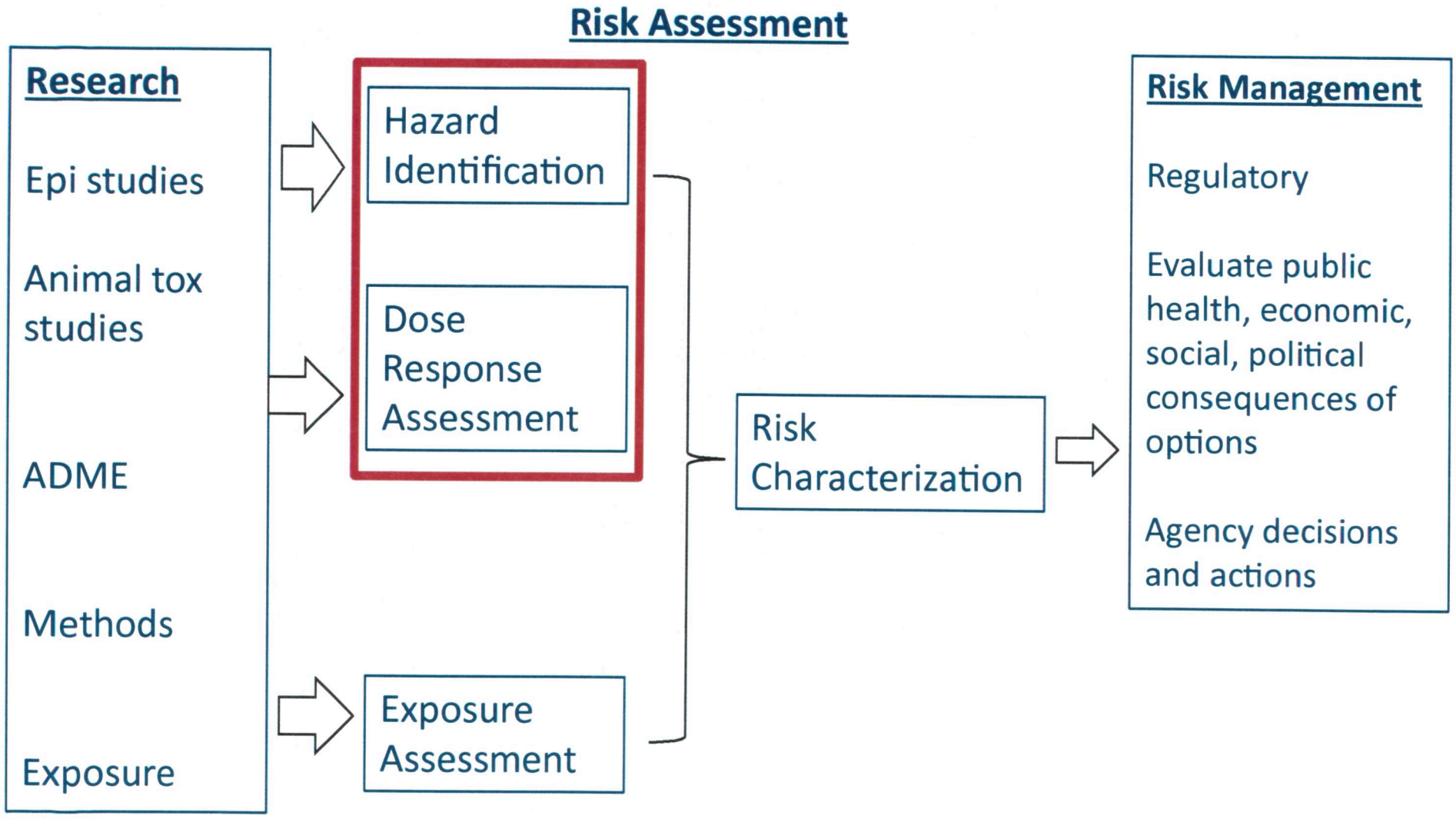
IRIS Program

- IRIS assessments critically review publicly available studies to:
  - –Identify adverse health effects
  - –Derive toxicity values



NRC risk assessment/risk management paradigm



Adapted from the National Research Council risk assessment risk management paradigm (NRC 1983).

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Hexavalent Chromium (Cr VI)

- Cr(VI) has been detected in drinking water throughout US
  - –Cr(VI) detected above 0.03 µg/L in ~75% of total samples (5260/6928)
  - –MRL:0.03 ppb; Most under ~5 ppb
    - Source: US EPA Third Unregulated Contaminant Monitoring Rule Occurrence data (as of October 2013)
    - EPA is continuing to compile data from public water systems and this only represents about 15 % of the data expected under the UCMR
- Cr(VI) reduces to trivalent chromium (Cr III) in biological fluids
  - –Cr(III) is poorly absorbed by cells, has limited toxicity
  - –Rodents chronically exposed to Cr(VI) via drinking water show toxicity and carcinogenicity in the GI tract

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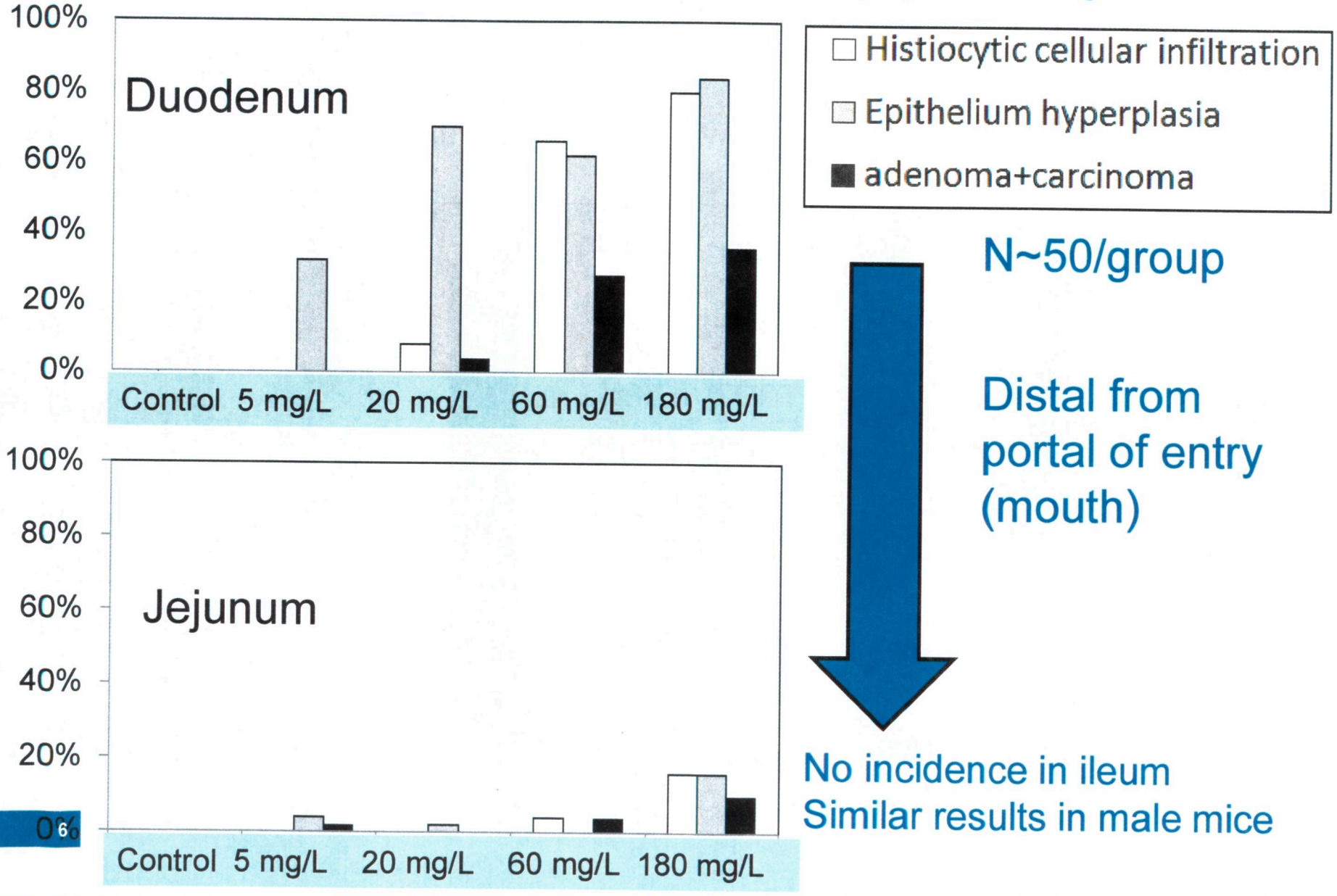
National Toxicology Program (2008)

Female **MICE** exposed for 2 years to Cr(VI) in drinking water



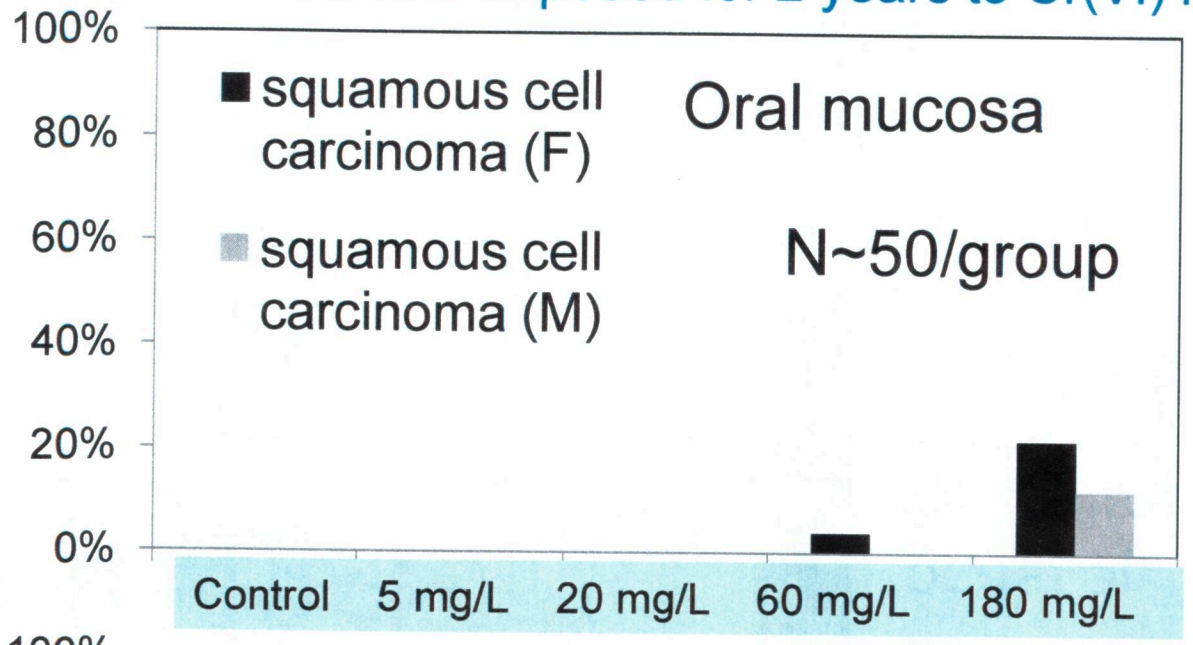
N~50/group

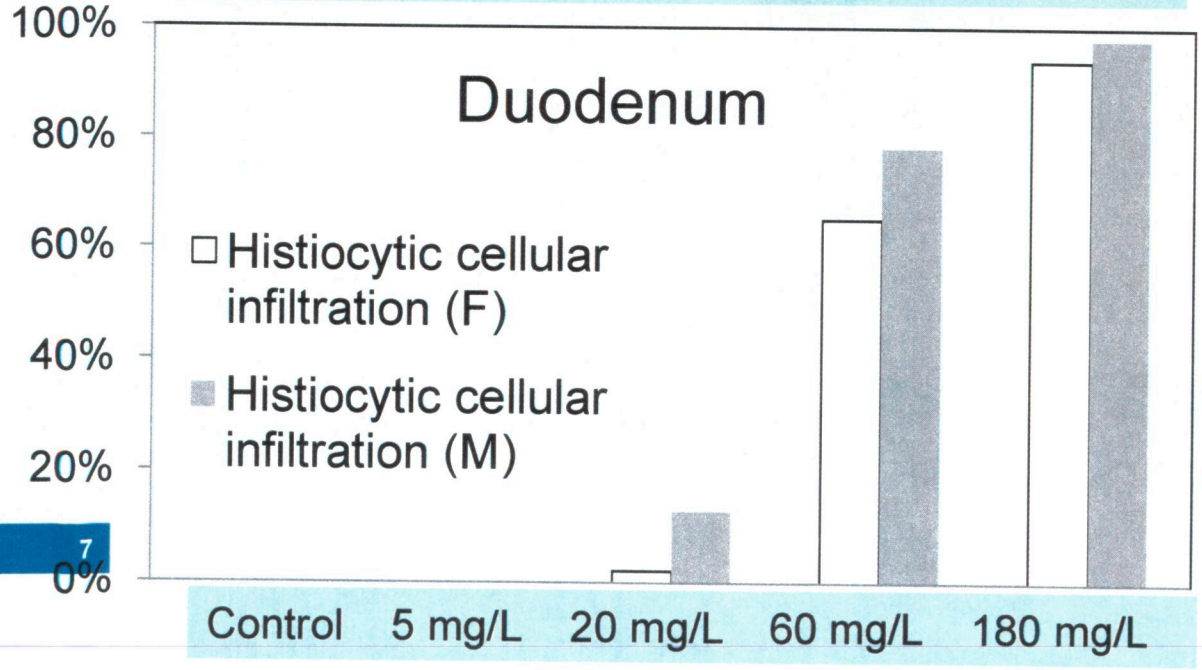
Distal from portal of entry (mouth)

No incidence in ileum
Similar results in male mice

National Toxicology Program (2008)

RATS exposed for 2 years to Cr(VI) in drinking water





Distal from portal of entry (mouth)

No tumors, no distal effects further along GI tract

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Evidence in humans is limited

- Zhang & Li (1987) and reanalysis (Beaumont et al., 2008)
  - Population in China chronically exposed to drinking water heavily contaminated with Cr(VI)
  - Currently the only study indicating elevated risk of stomach cancer in humans

- IARC determined this single study was insufficient to constitute evidence of an association between oral exposure to Cr(VI) and stomach cancer
  - **International Agency for Research on Cancer** (2012). IARC Monographs: *A review of human carcinogens: Arsenic, metals, fibres, and dusts.*

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Physiologically-based pharmacokinetic (PBPK) modeling

- Modeling tools that can help explain similarities and differences in response between species
- Can be used to extrapolate animal results to humans
- Can aid in modeling Cr(VI) reduction in vivo

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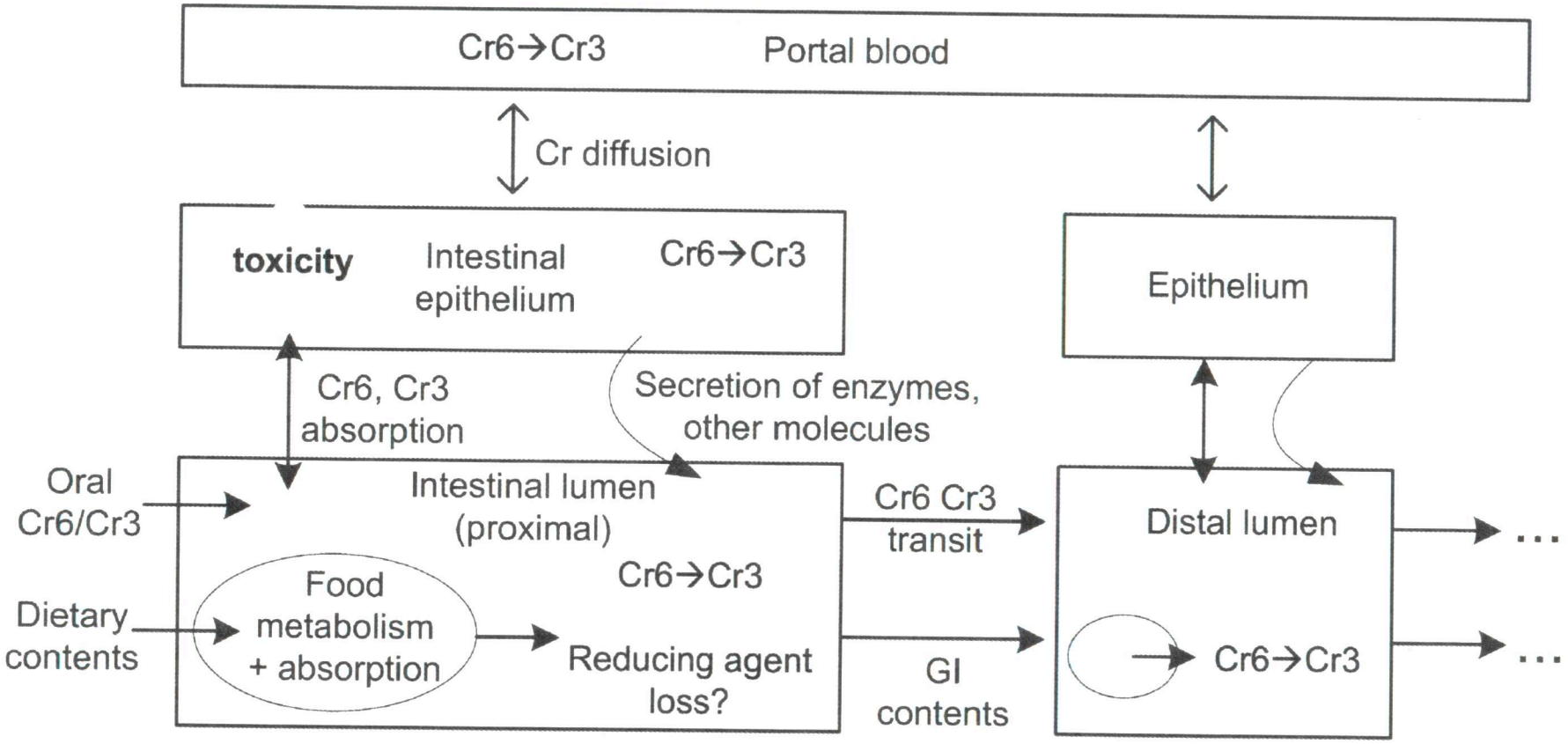
Measurement complexities

- Only possible to analytically measure **total** chromium *in vivo*
  - –Total chromium = Cr(VI) + Cr(III)

- Oral ingestion of Cr(VI) leads to absorption of a Cr(VI)/Cr(III) **mixture** due to reduction
  - –Difficult to know which form passes through the intestine
  - –High red blood cell (RBC) to plasma ratios may indicate Cr(VI) uptake: RBCs rapidly absorb and reduce Cr(VI), "trapping" Cr(III)

- Dietary exposure to Cr(III) occurs in all species

Competing transport, reduction, and uptake



- Higher **total** chromium in body following Cr(VI) exposure, compared to Cr(III) exposure (NTP, 2008, 2010)
- Tissue chromium concentrations decrease distally (Kirman et al., 2012): **duodenum** > jejunum > ileum

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PBPK models of chromium

- O'Flaherty (1996) PBPK model in rats
  - –Calibrated to data from intravenous, gavage, inhalation, and drinking water (pre-1985 data)
  - –Insufficient model for GI tract kinetics
  - –Incorporated background Cr(III) exposure
- Kirman et al. (2012) PBPK model in rats and mice
  - –Calibrated with new data, but drinking water studies only
  - –Complex model for GI kinetics
  - –Neglected background Cr(III) (subtracted concentrations of control from the exposure data)
- **This work attempts to reconcile differences and incorporate best science from both models**

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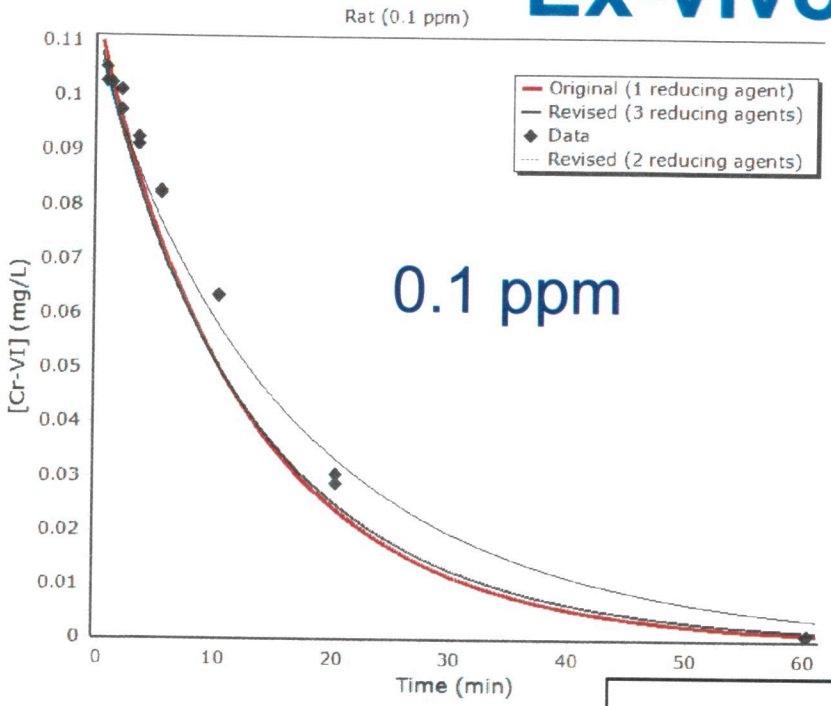
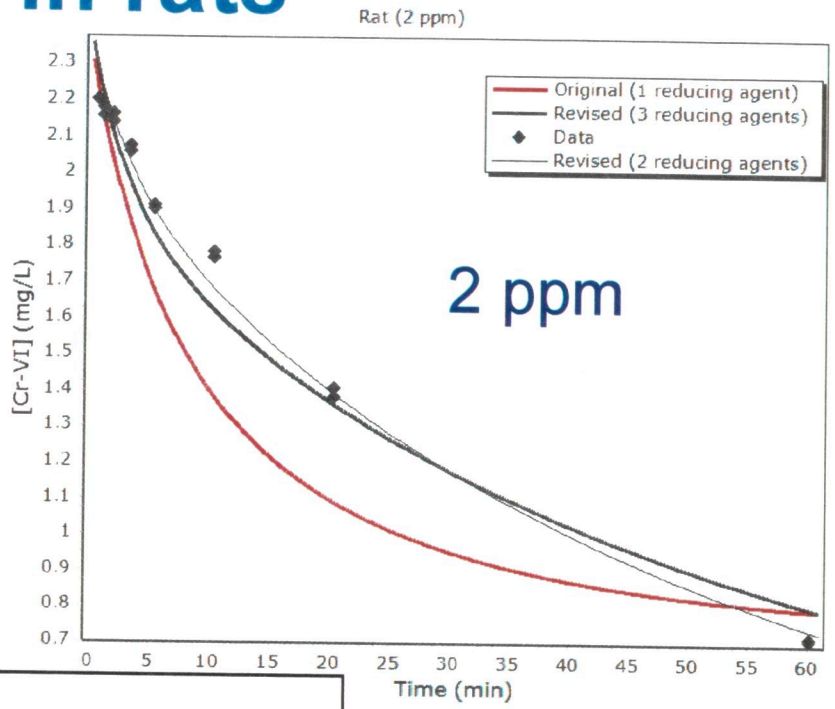
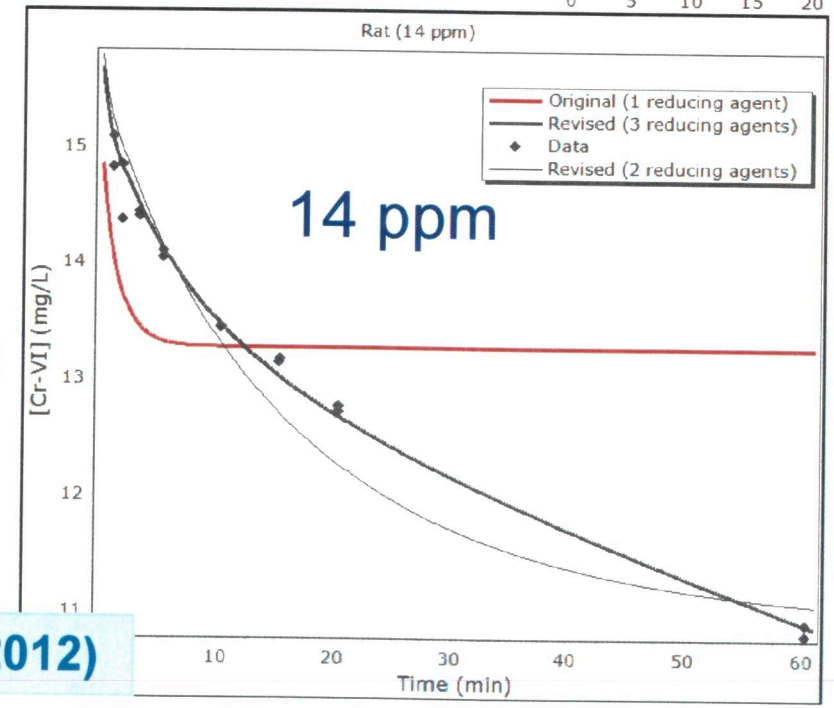
Revision of GI kinetics

- **Original assumption:** One lumped component of the gastric juice is capable of reducing Cr(VI) to Cr(III)
  - –Once this reducing agent is depleted (i.e., at high Cr(VI) levels) no more reduction can occur
- **Alternative assumption:** Two or three reducing agents exist in the gastric juice, each with different kinetic rates and capacities to reduce Cr(VI)
- These assumptions have implications when interpreting toxicological data
  - –How much un-reduced Cr(VI) gets into the body?
  - –What are the species differences?

Ex-vivo data in rats



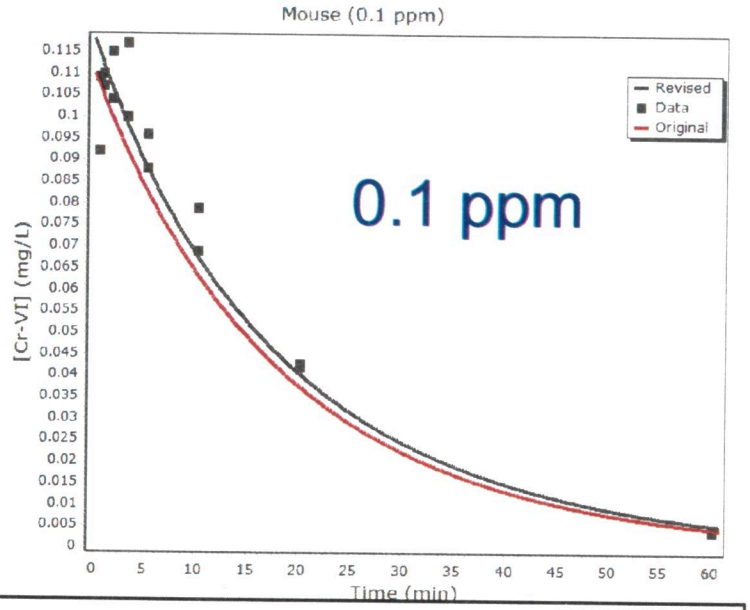


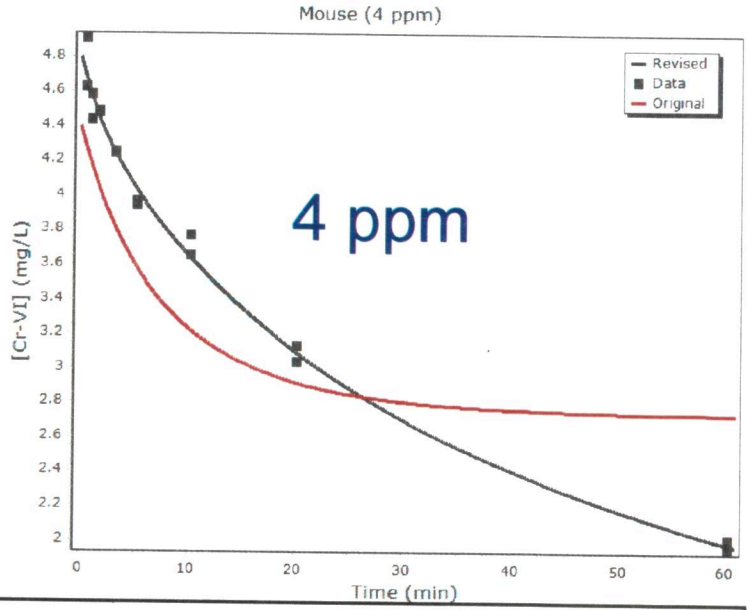


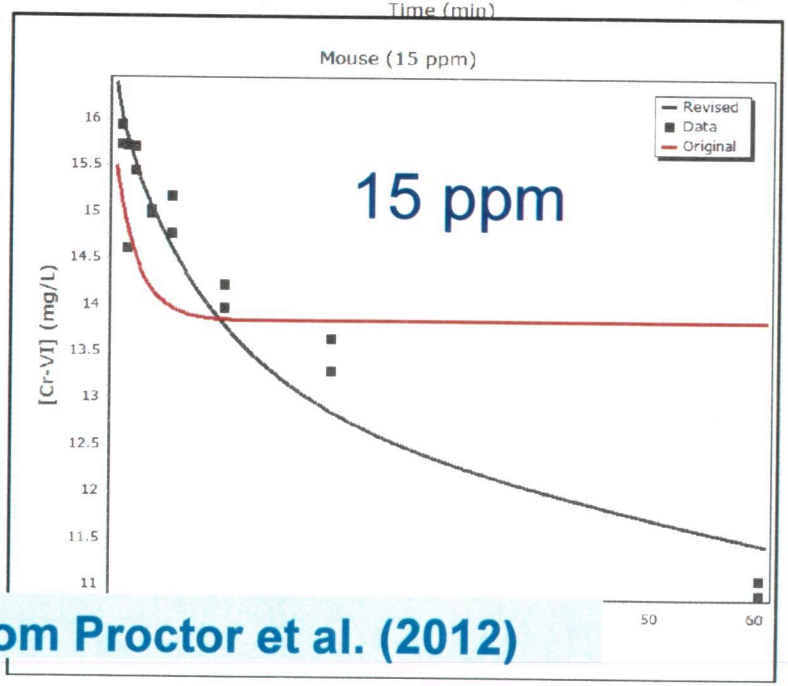
Original model fails at high concentration in gastric juice

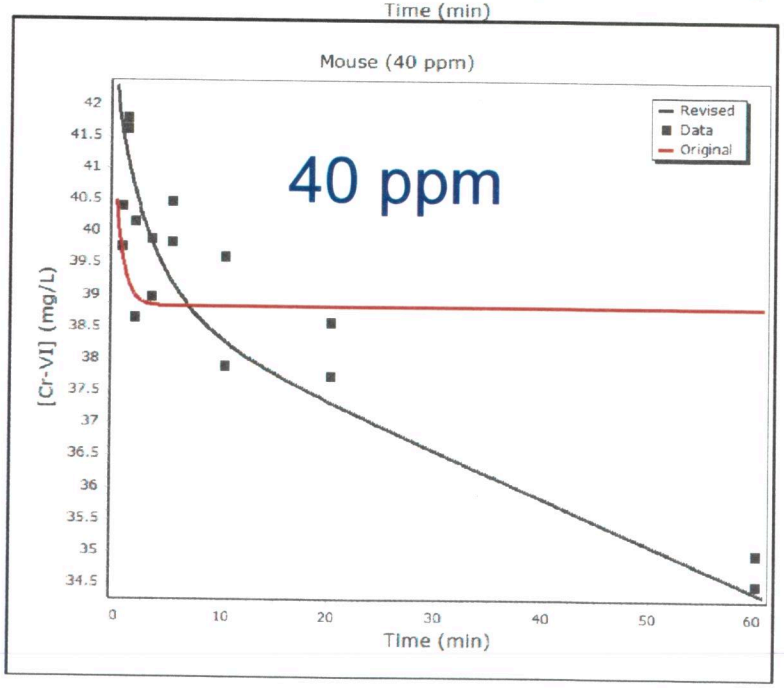
Data from Proctor et al. (2012)

Ex-vivo data in mice





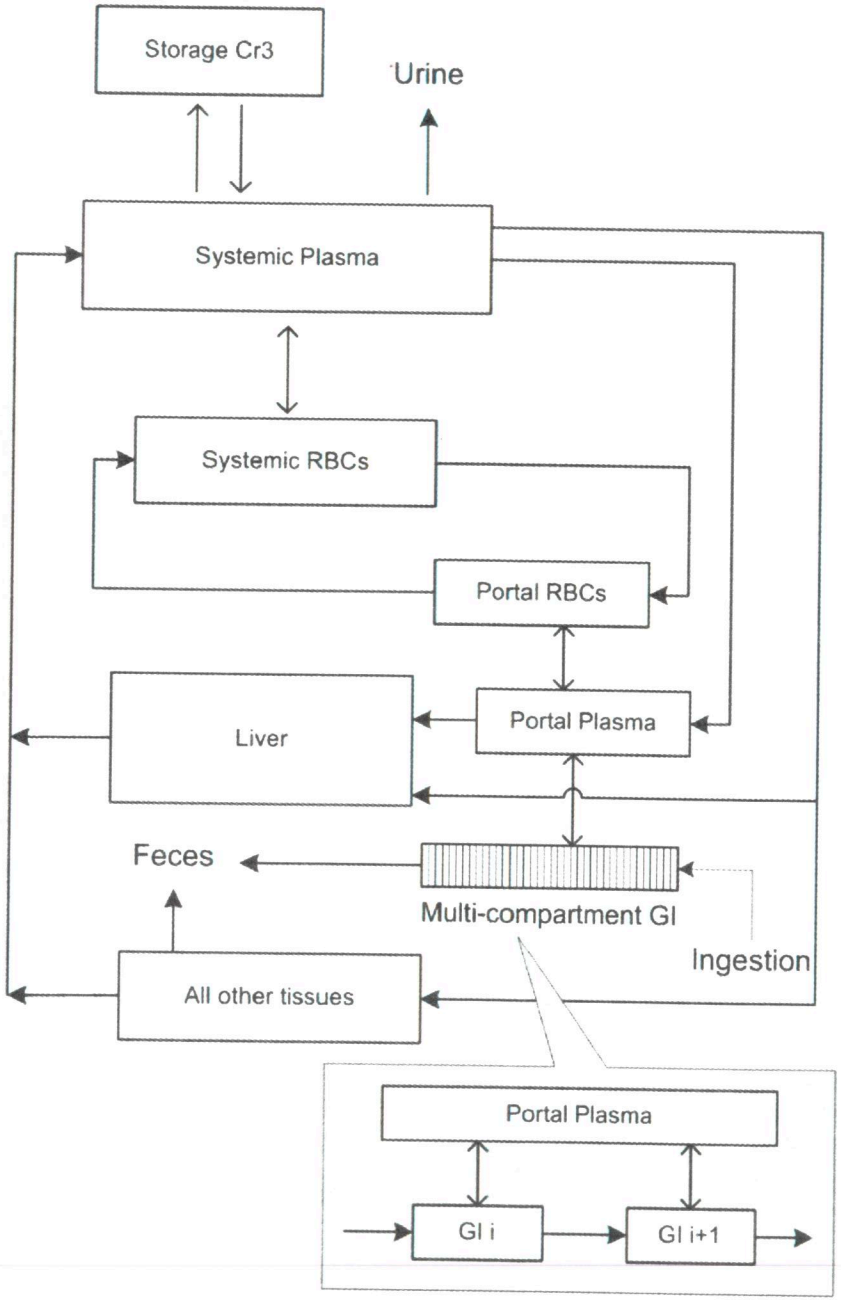




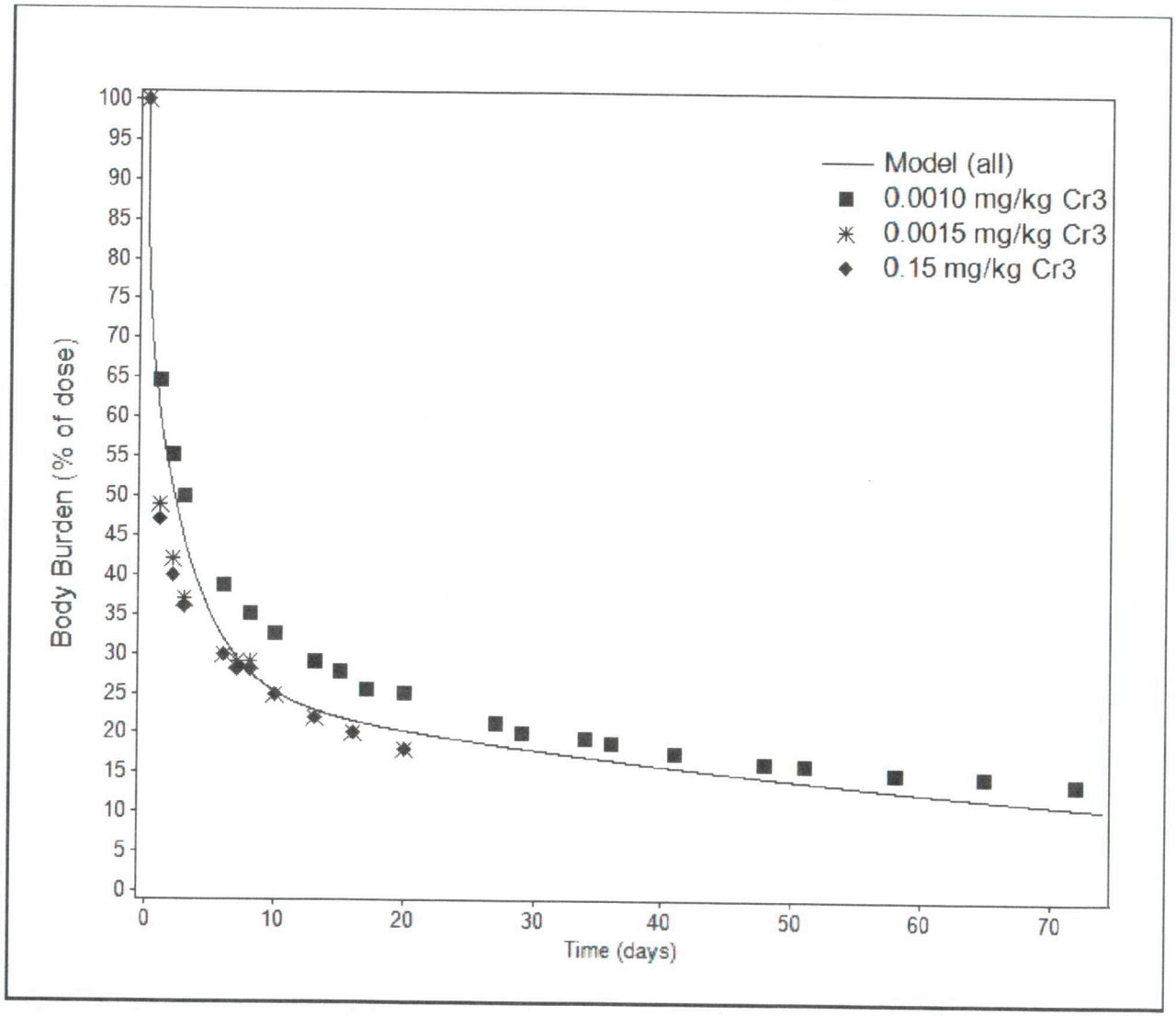
Data from Proctor et al. (2012)

Revised PBPK model

- Adapted from the model by Kirman et al. (2012)
- Revised GI kinetic model
- Simplified whole-body kinetics
  - –More focus on GI, and total body burden
- Attempt to fit intravenous, gavage, and drinking water routes with consistent parameters
  - –Incorporate background Cr(III) exposure in chronic studies

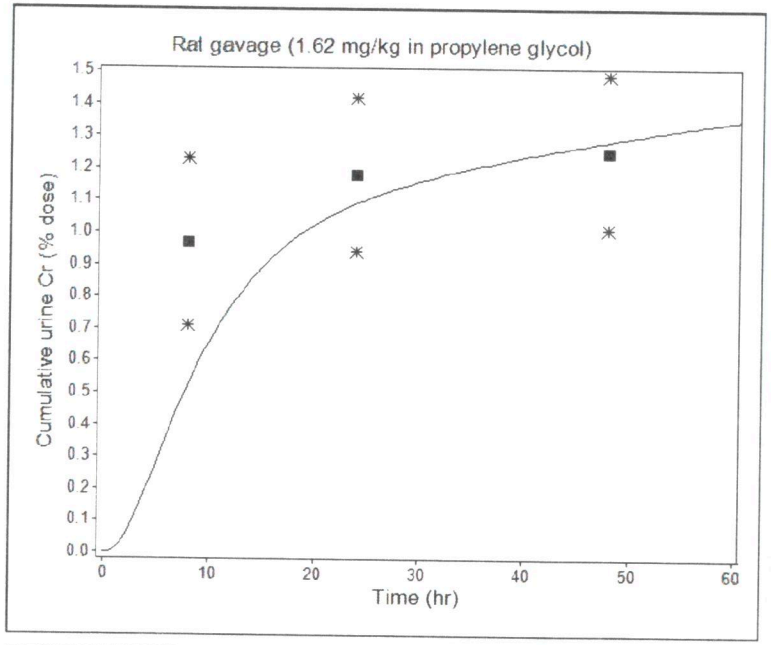


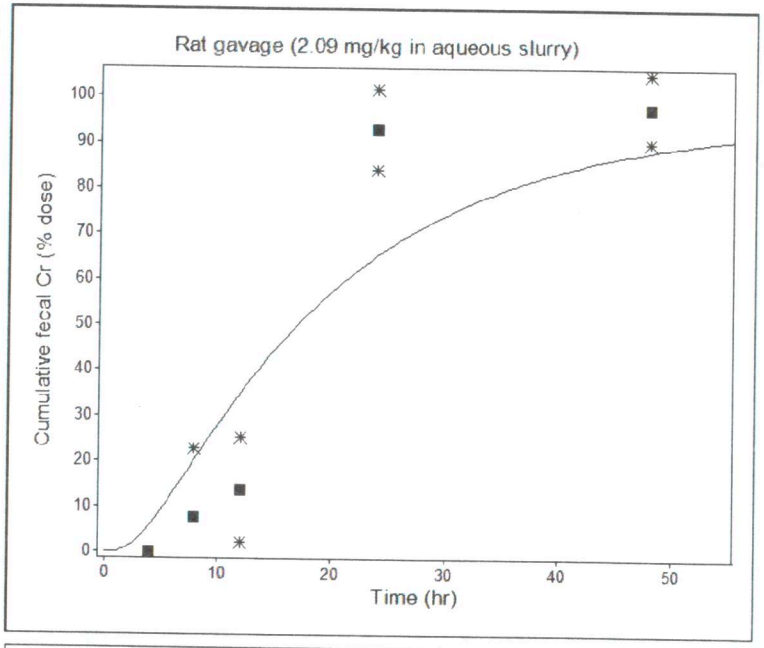
Intravenous data

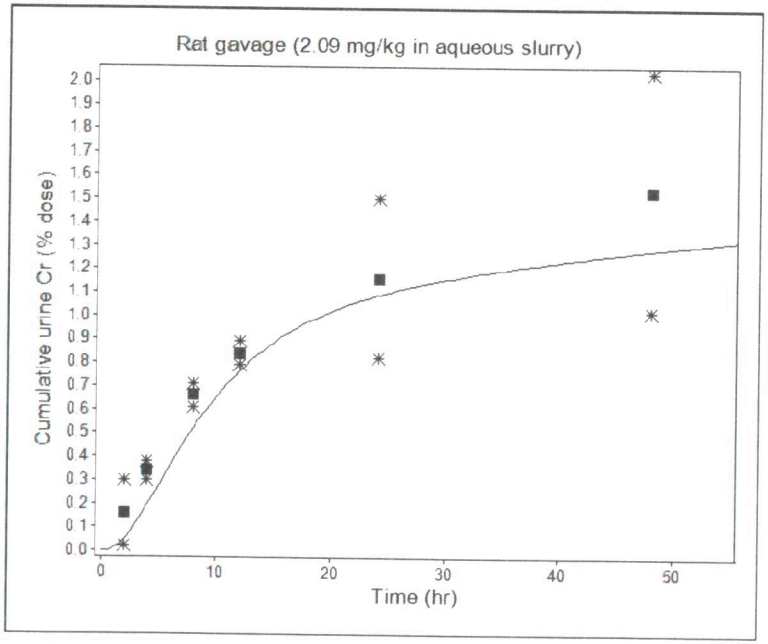


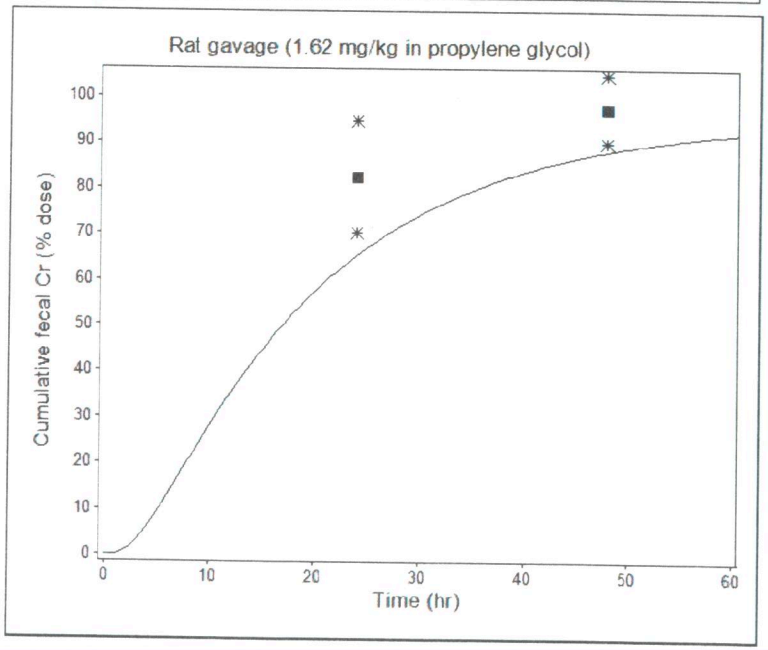
Parameters for long-term retention and urinary elimination were fit to data by Mertz et al. (1965) (above) and Sayato et al. (1980)

Rat gavage Cr(III) data from NTP (2010)

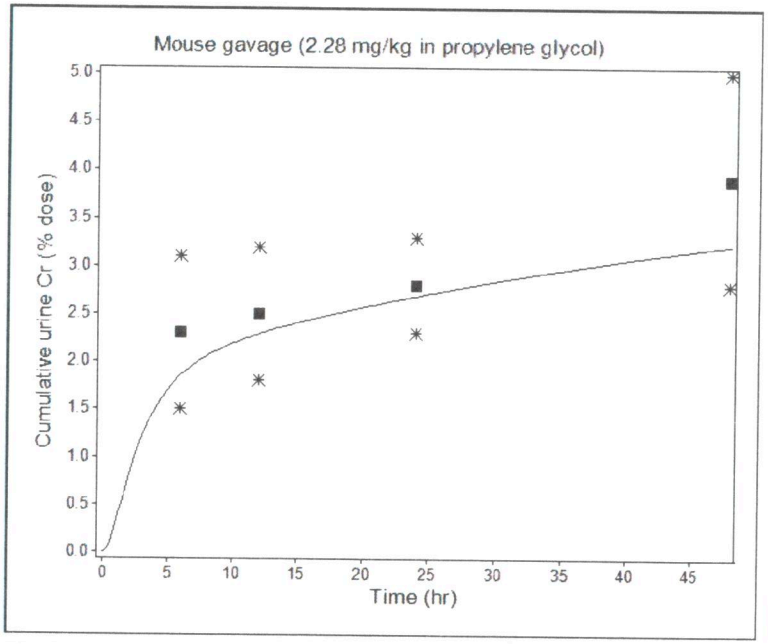


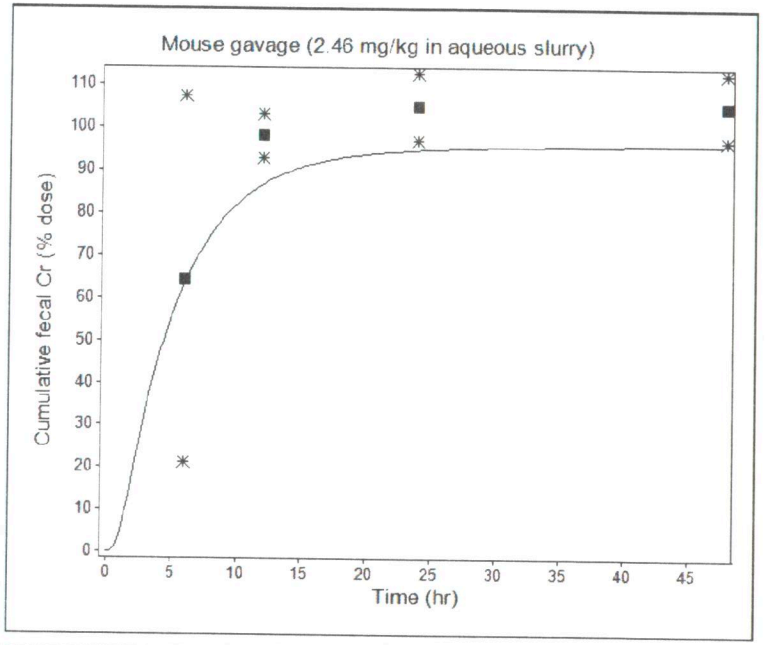


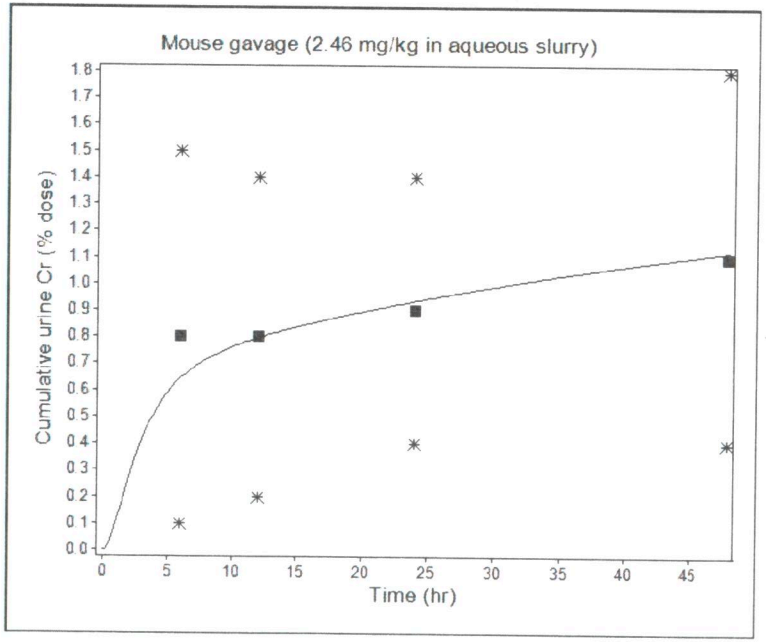


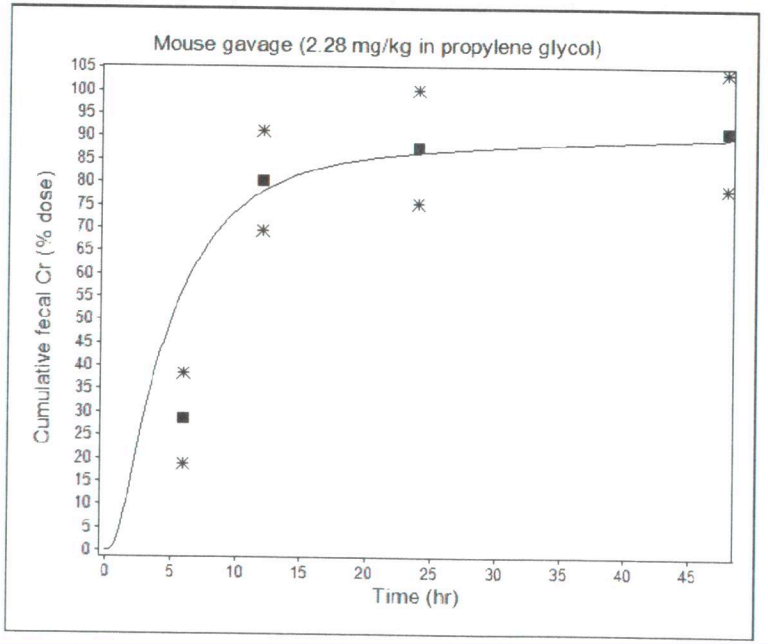


Mouse gavage Cr(III) data from NTP (2010)

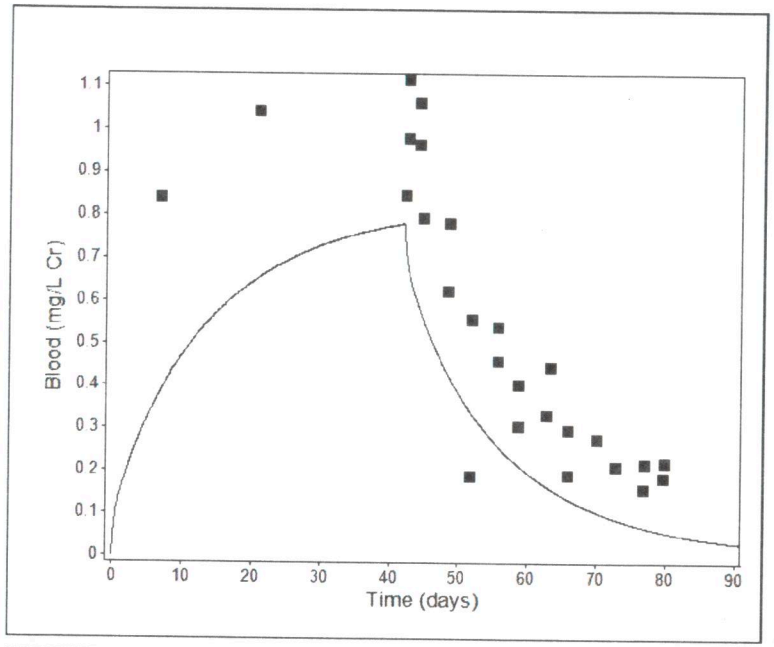


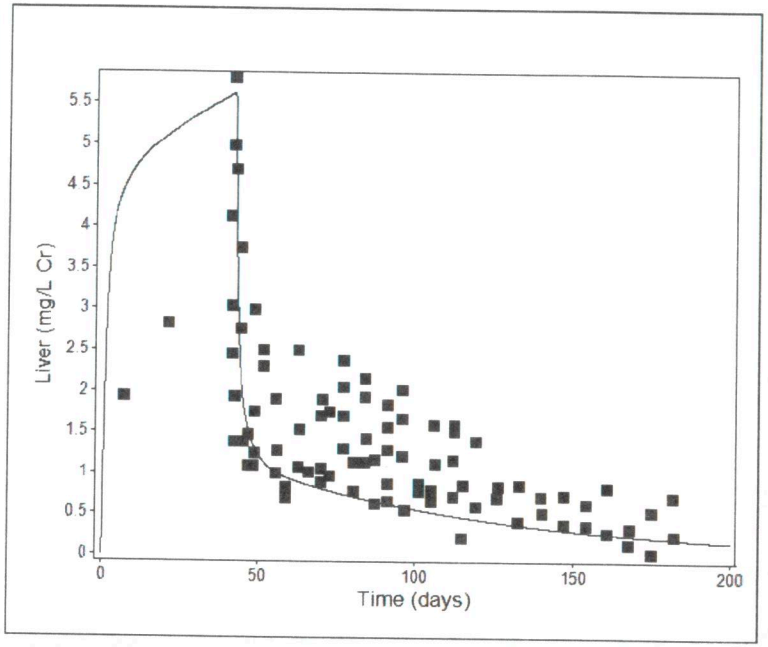


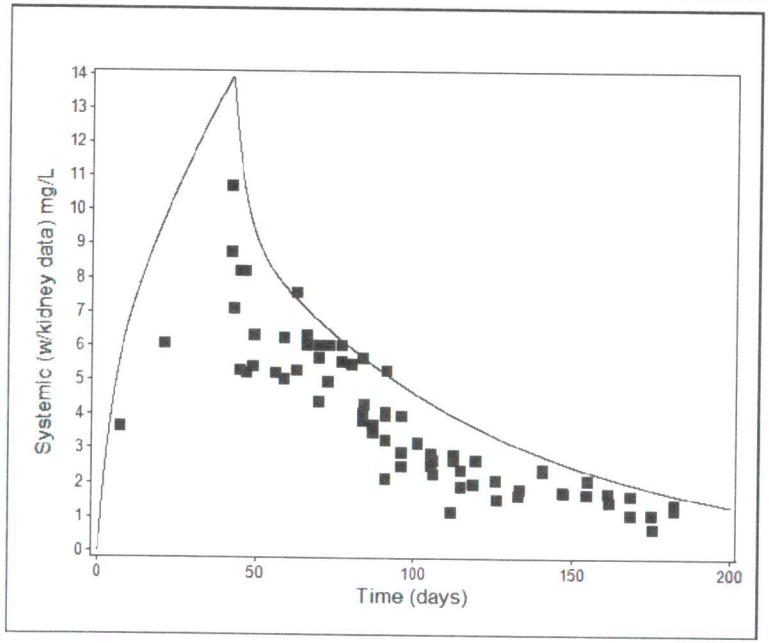




Rat data fromThomann et al. (1994)



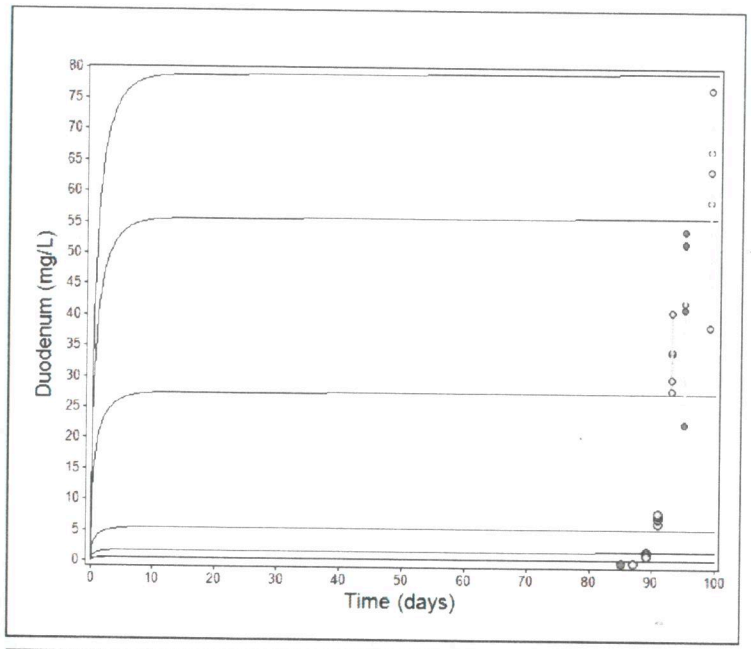


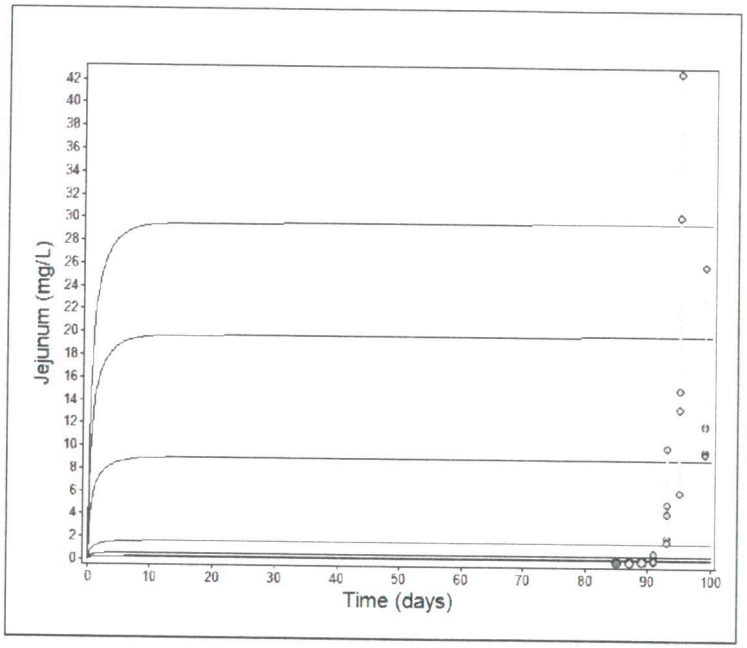


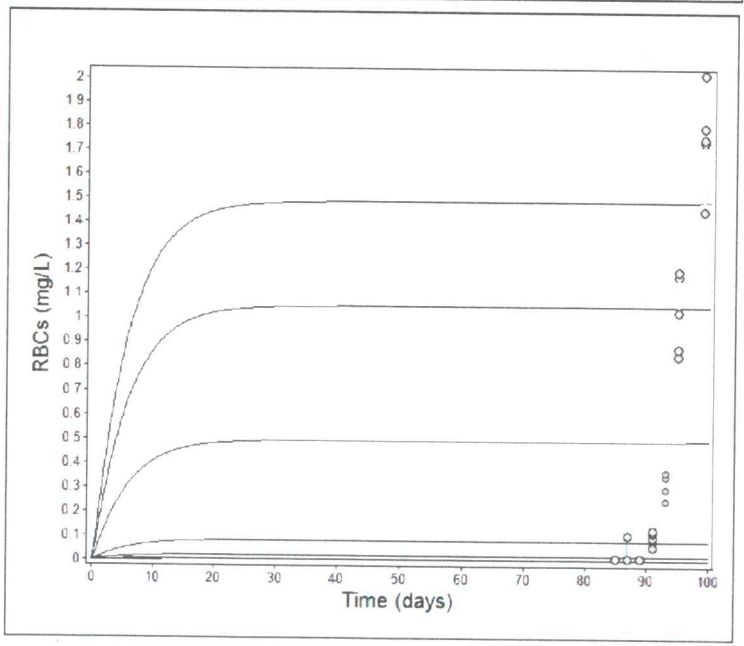
Rats exposed to Cr(VI) in drinking water at 100 mg/L for 42 days, followed by ~100 days without exposure

Data points represent individual rat measurements [digitized by the study authors of Kirman et al. (2012)]

Mouse data from Kirman et al. (2012)







Mice exposed to Cr(VI) in drinking water at varying doses for 90 days.

Data points represent individual rat measurements [provided by Kirman et al. (2012)]

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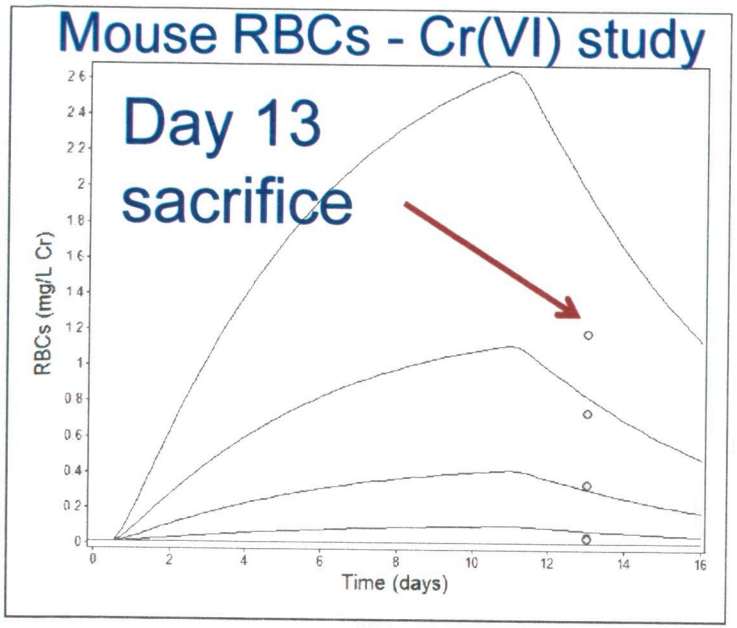
Data also adequately fit to:

- Kargacin et al. (1993) chronic drinking water data in rats and mice (liver, blood, lumped systemic tissues)

Additional modeling revisions:

- Added uptake of Cr(III) into GI tissues via plasma perfusion
- Most parameters are identical for both rats and mice
- All parameters are the same for all data sets
  - With exception of GI absorption, which is expected to vary with formulation and study

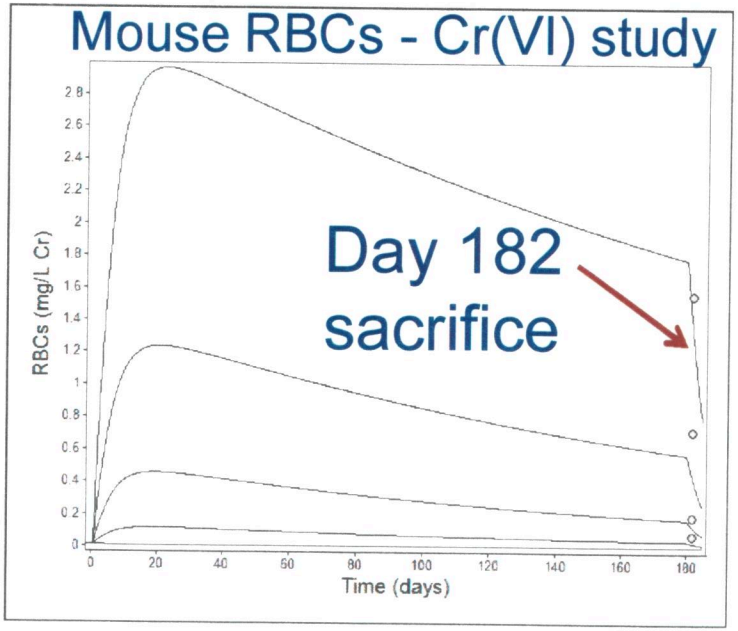
PBPK modeling of the NTP data

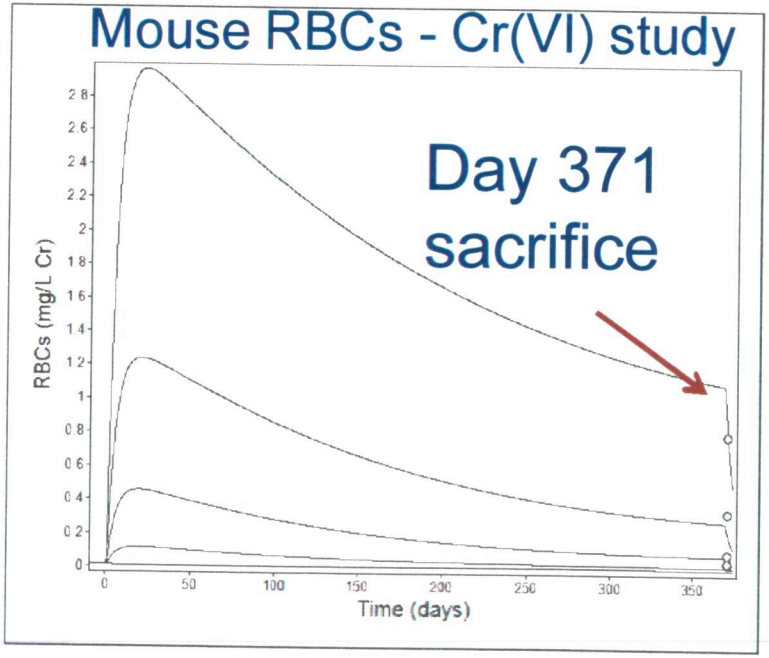


Challenges

- Rapid body weight and dose change
- 48-hour "wash-out" period
- Sacrifice time vs. final Cr(VI) dose
- Background Cr(III) exposure

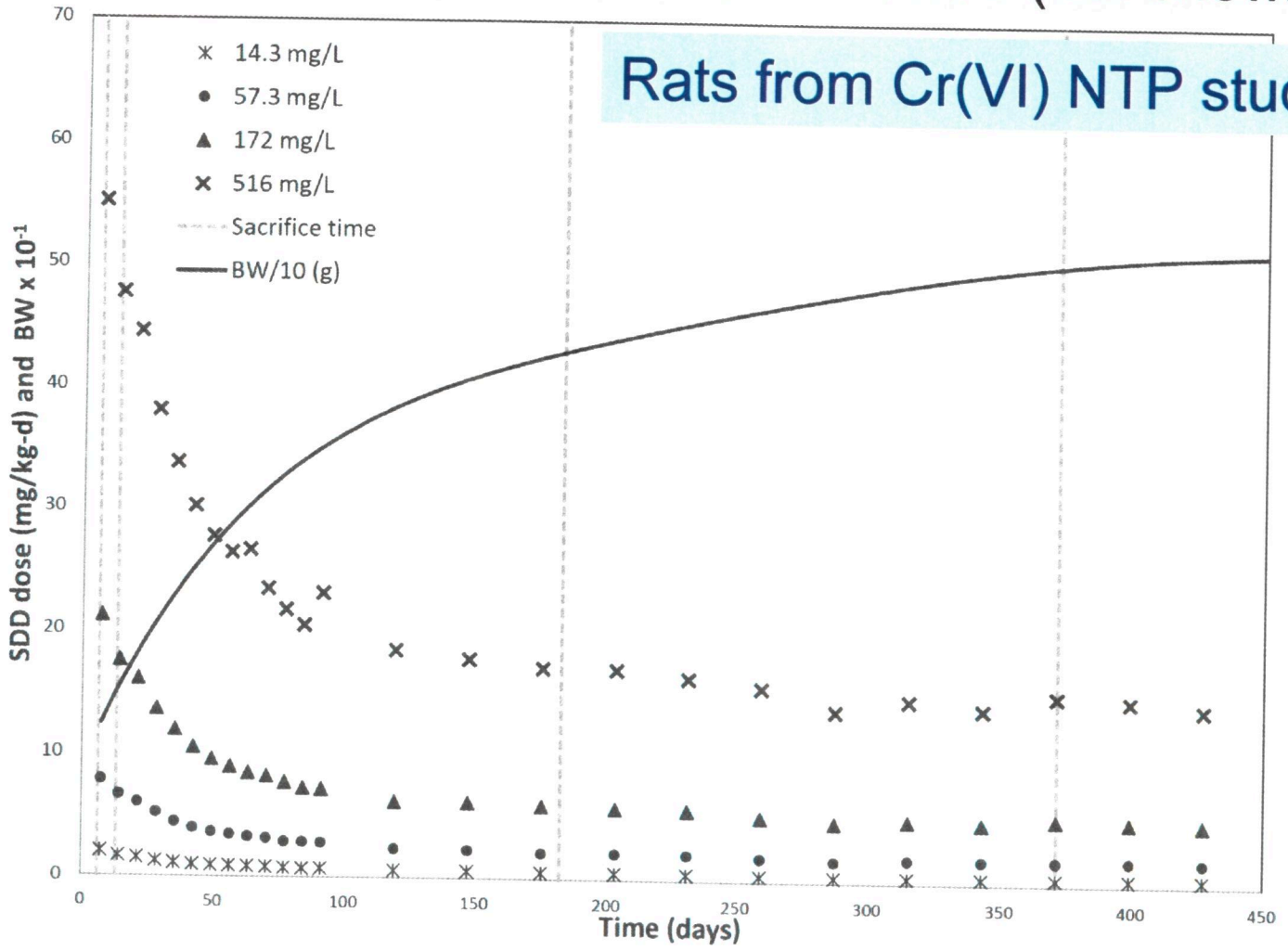
Chronic NTP data used primarily as a validation



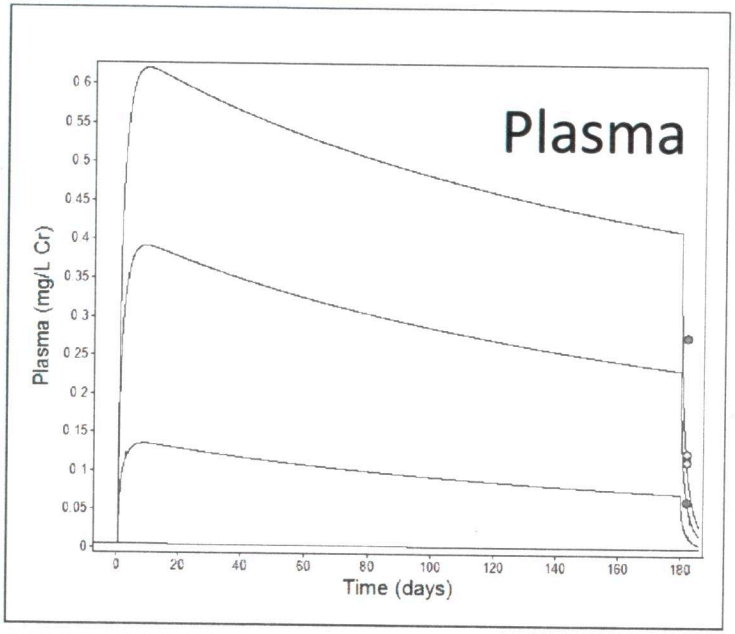


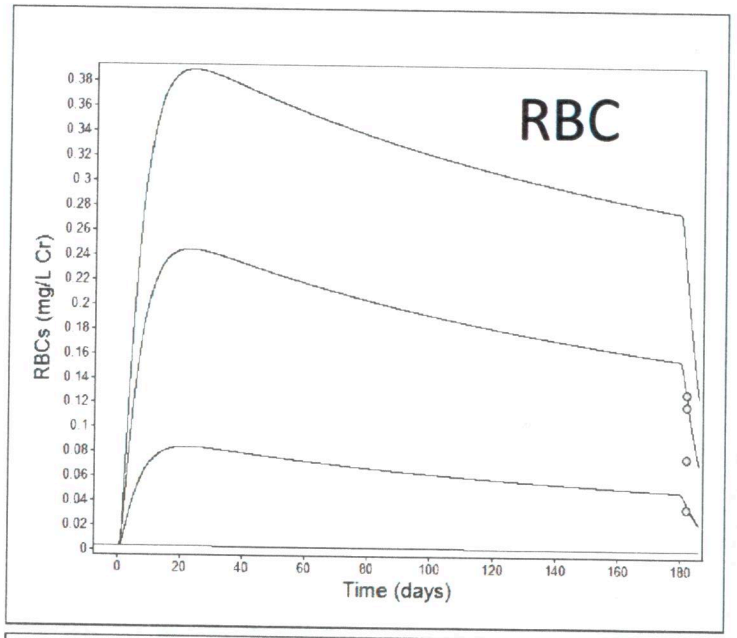
Body weight and dose curves

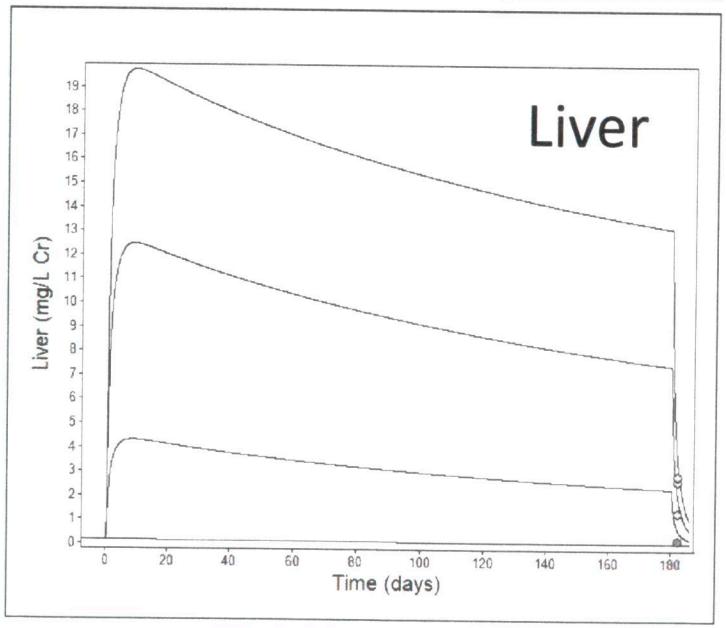
- Drinking water rate (thus mg/kg-d) and body weight functions incorporated into model
- High dose groups had different BW curve (not shown)

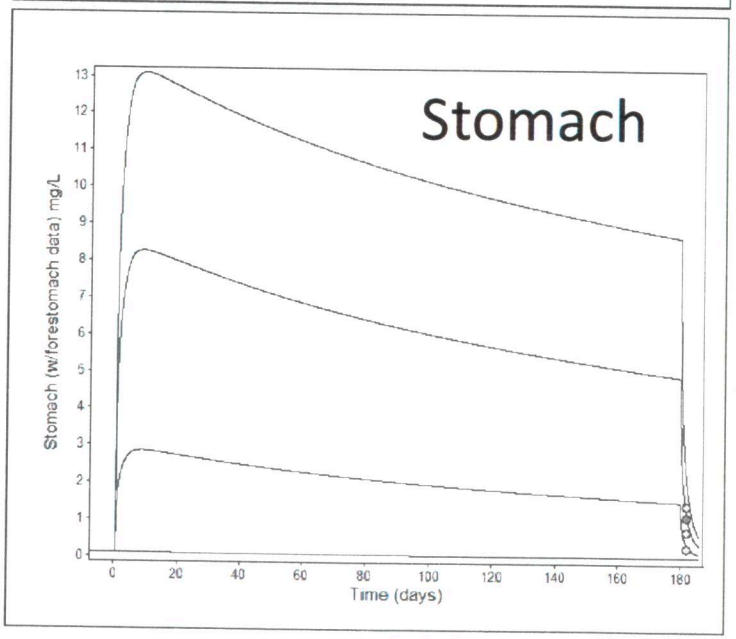


Dietary Cr(III) from NTP (mice): Day 182 sacrifice

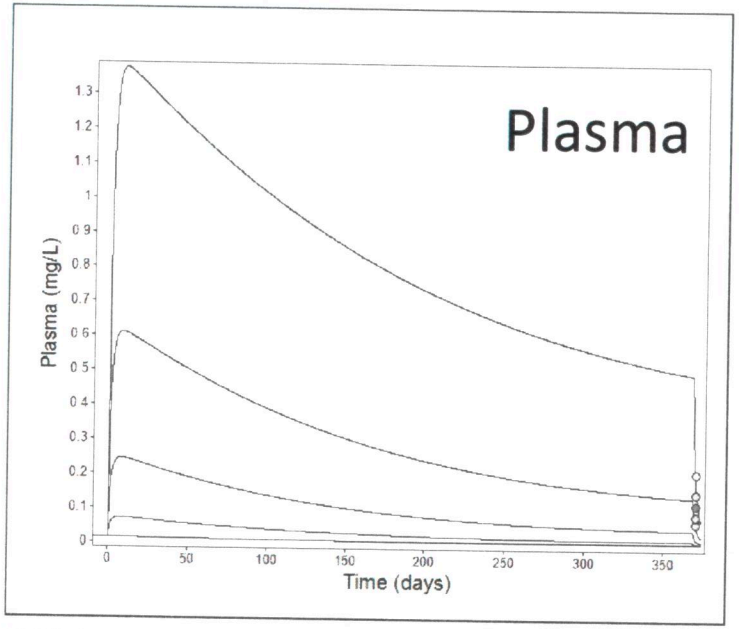


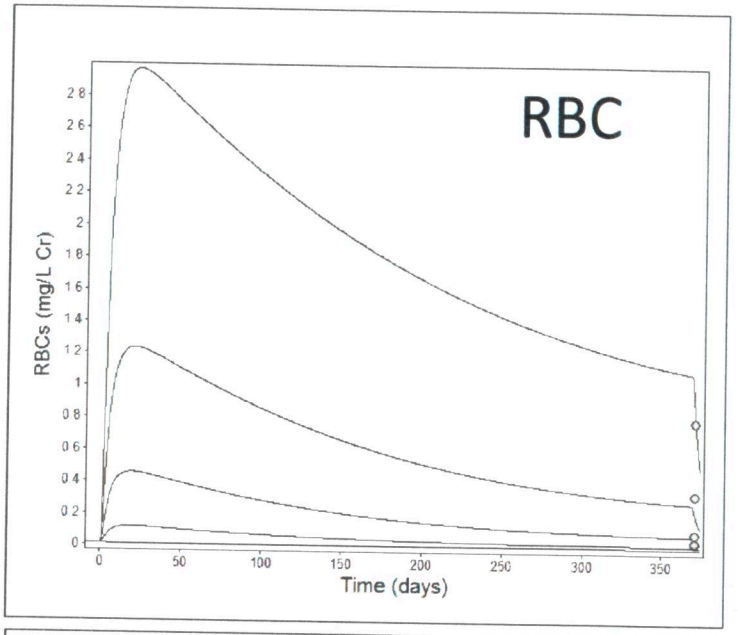


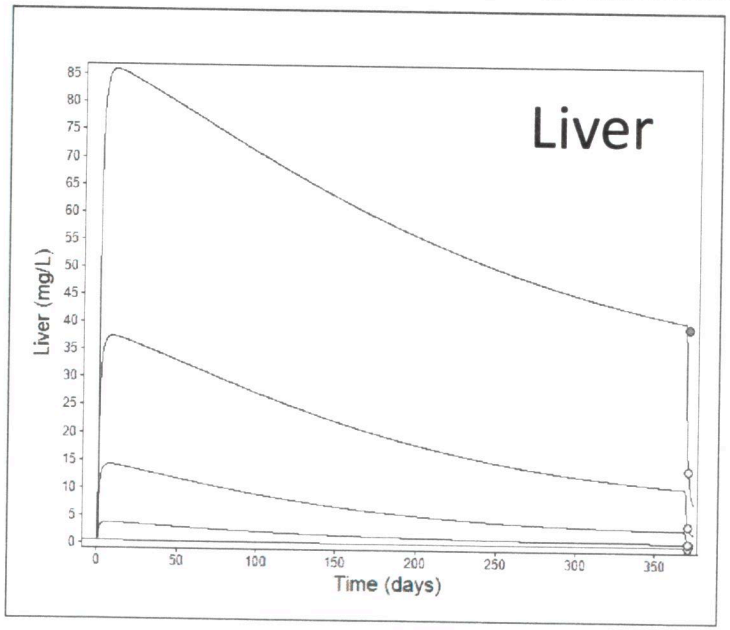


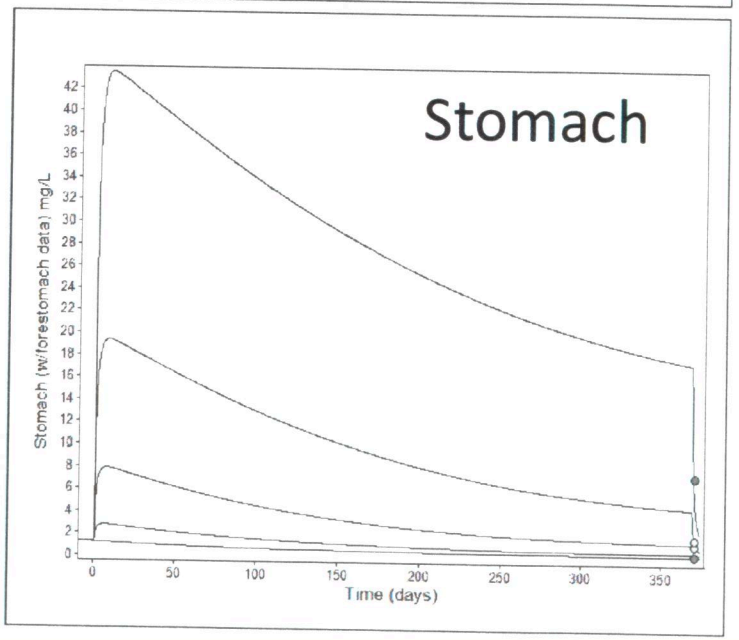


Dietary Cr(VI) from NTP (mice): Day 371 sacrifice









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Unanswered questions

- Uncertainties that go beyond idealized GI kinetics
  - –Time variation in GI motility and secretions
  - –Inter-individual variability in GI transporters for Cr(VI)
  - –Variation of transporters along GI tract
  - –Variation in diet and nutrition
  - –Age susceptibilities
  - –What about sites upstream of stomach?
    - Oral cavity, tongue, esophagus

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Unanswered questions

- Best internal dose-metric for GI tract toxicity?
  - –Amount of Cr(VI) absorbed
  - –Amount of Cr(VI) escaping reduction in the stomach
  - –Concentration of Cr(VI) in sensitive GI compartments
  - –Rate of reduction (i.e., ROS generation) at sensitive sites

- Cr(VI) reduction webinar (Sep. 19 & 25 of this year)
  - –Talks and discussions from many perspectives
  - –Materials available at:

    http://www.epa.gov/iris/irisworkshops/cr6

Acknowledgements

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Lynn Flowers

External

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Cr(VI) Webinar team

Audrey Turley
Courtney Skuce
Maureen Johnson (EPA)

Panelists

Elaina Kenyon (EPA)
Gary Ginsberg
Kim Barrett
Max Costa
John Crison
Silvio De Flora
Sean Hays

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Summary

- A harmonized PBPK model was developed for rodents
  - –Adapted model by Kirman et al. (2012) to incorporate revised GI kinetics
  - –Incorporated some features of O'Flaherty (1996) model
  - –Model re-fit to data from additional routes of exposure
  - –A model for humans is also under development
- Model will aid the evaluation of dose-response data for the IRIS Toxicological Review of Hexavalent Chromium

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